Supporting Information

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A

oligonucleotide	expected mass (Da)	observed mass (Da)	Note
Oligo conjugated to α-NCEH1, fig. 3D	10323.98	10327.75	
Oligo conjugated to SA, fig. 3D	11340.55	11348.61	
Oligo 1 for α-GAPDH, fig. 4A	12829.15	12836.66	
Oligo 2 for α-GAPDH, fig. 4A	18947.35	18966.92	+Na

B

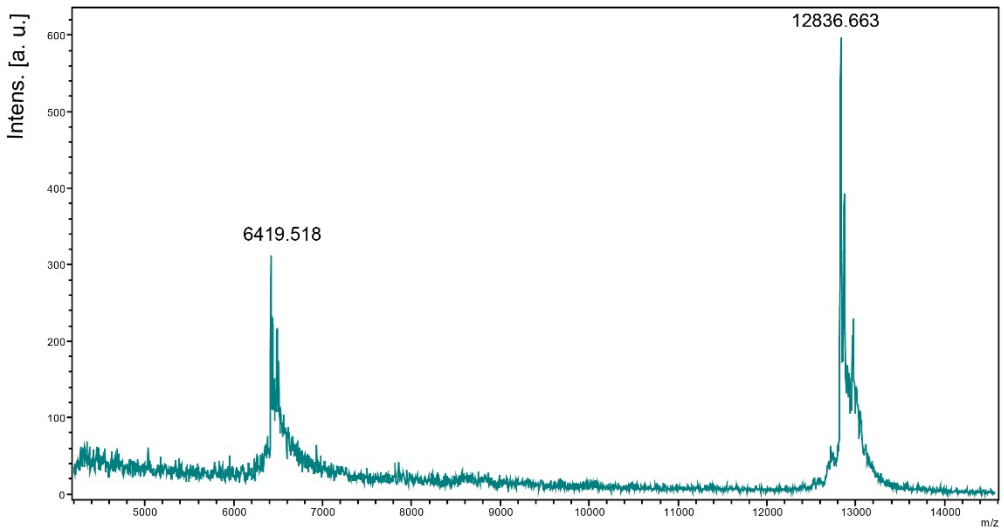

Figure S1 (A) characterization of succinimidyl-modified oligonucleotides used in figure 3 and figure 4. (B) One representative MALDI-TOF spectrum for succinimidyl-modified oligonucleotide conjugated to poly α-GAPDH antibody.

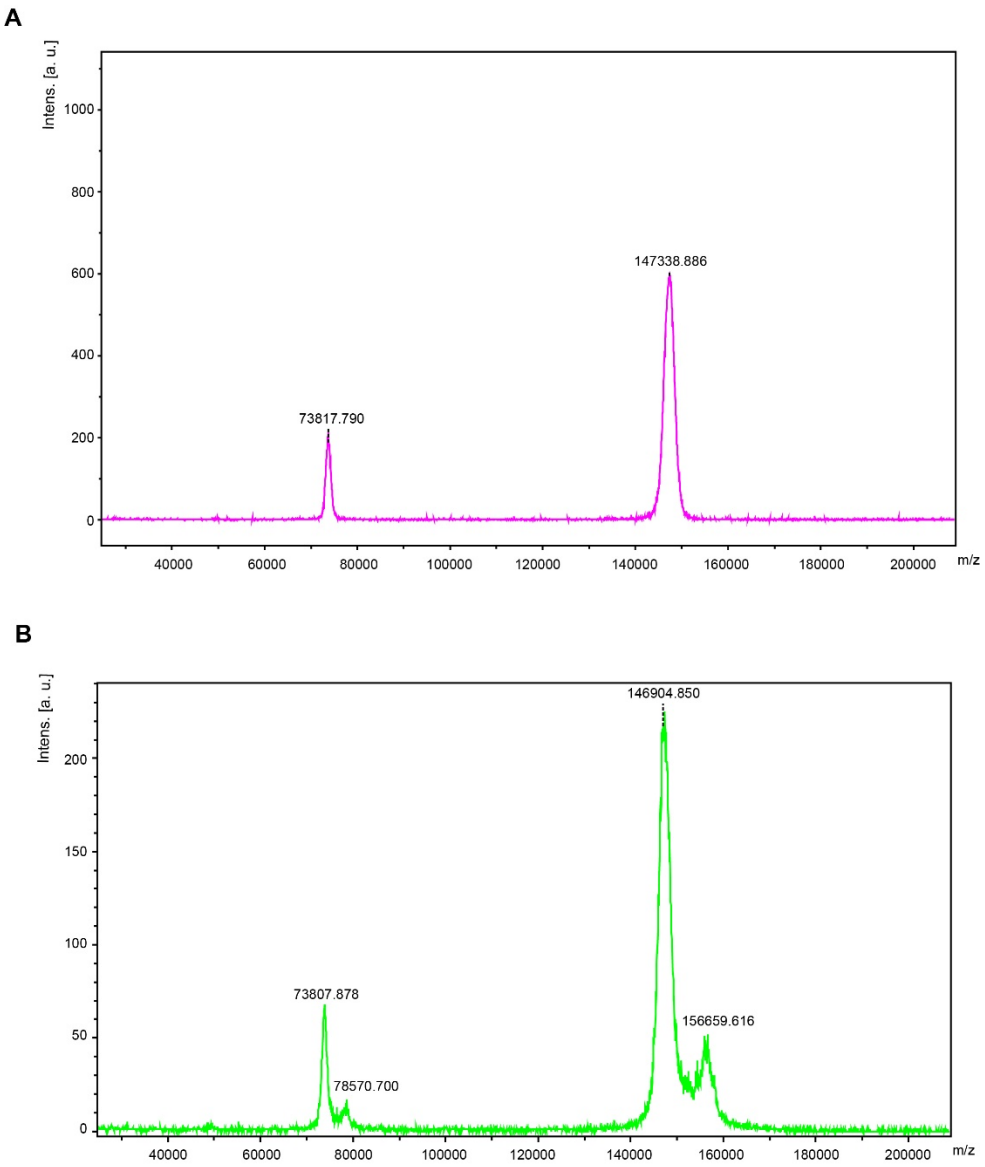

Figure S2 Characterization of antibody-oligonucleotide conjugates by MALDI-TOF. (A) representative MALDI-TOF spectrum of pure IgG. (B) representative MALDI-TOF for oligonucleotide conjugated IgG. The molecular weight of the oligonucleotide is 10224 Da.

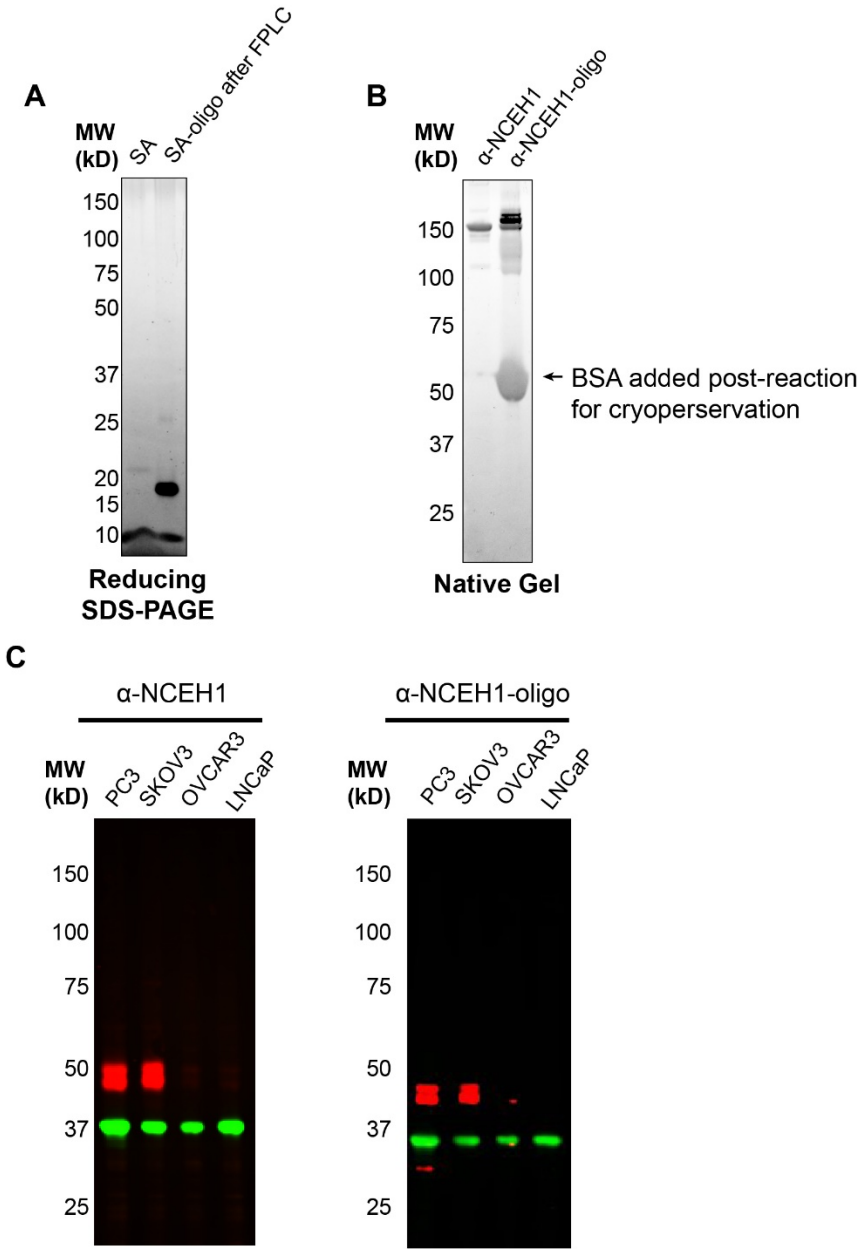

Figure S3 (A) Reducing SDS-PAGE gel validation of streptavidin conjugated oligonucleotide after FPLC purification involved in figure 3. (B) Native PAGE gel validation of α-NCEH1 antibody conjugated with oligonucleotide involved in figure 3. BSA was added after the conjugation as a component of storage buffer. (C) Comparison of the NCEH1 antibody activity and specificity before and after oligonucleotide conjugation. PC3 and SKOV3 cell lysate have high NCEH1 expression whereas OVCAR3 and LNCaP have low expression. The green bands at about 37 kD are the GAPDH loading control.

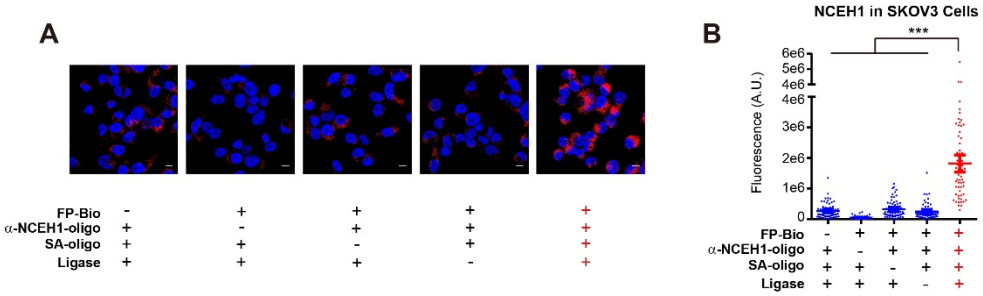

Figure S4 (A) ADPL with directed conjugated probes to detect the NCEH1 activity in SKOV3 cells in the presence or absence of the indicated ADPL components. Channels shown are DAPI nuclear stain (blue), ADPL signal (red). (B) quantification of ADPL signal in the presence or absence of the indicated ADPL components, demonstrating the probe-, ligation- and POI- dependent nature of a robust ADPL signal using the direct conjugated probe.

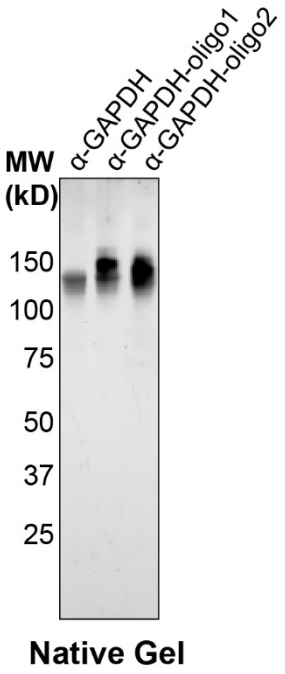

Figure S5 Native PAGE gel validation of polyclonal α-GAPDH antibody conjugated with oligonucleotide 1 and oligonucleotide 2.

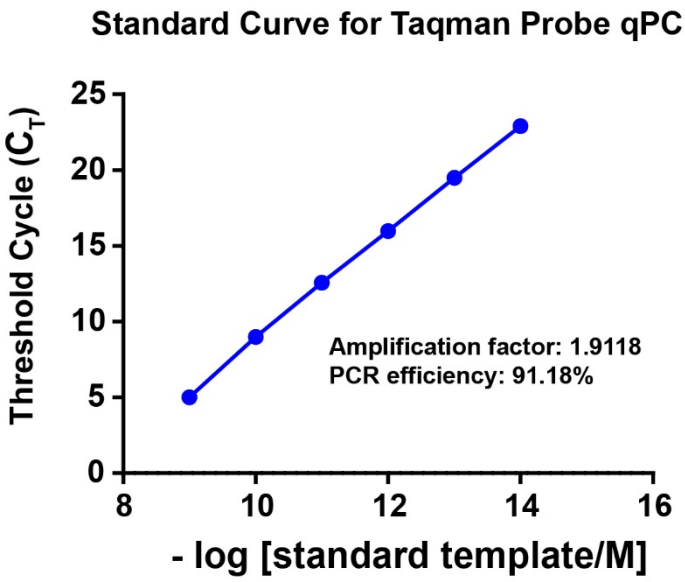

Figure S6 Standard curve for Taqman probe based qPCR. The derived amplification factor was used to convert the C_T difference to fold change.

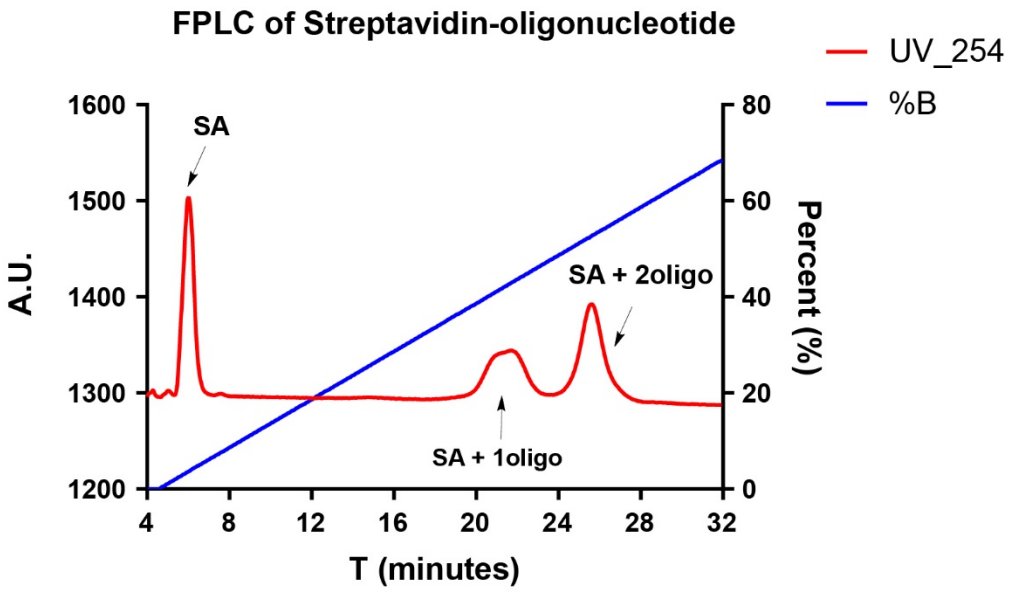

Figure S7 FPLC chromatogram for purification of streptavidin-oligonucleotide conjugate.

Table S1: Oligonucleotides involved in Figure 3

Oligo names	Sequence
Oligo conjugated to α-NCEH1	5'-amine-AAAAAAAAAATATGACAGAACTAGACACTCTT-3'
Oligo conjugated to streptavidin	5'-amine-AAAAAAAAAAGACGCTAATAGTTAAGACGCTTmUmUmU-3'
Short bridging oligonucleotide	5'-OPO_3-GTTCTGTCATA TTTAAGCGTCTTAA-3'
Long bridging oligonucleotide	5'-OPO_3-CTATTAGCGTCCAGTGAATGCGAGTCCGTCTAAGAGAGTAGTACAGCAGCCGTCAAGAGTGTCTA-3'
Detecting oligonucleotide	5'- Alexa 555-CAGTGAATGCGAGTCCGTCT-3'

Table S2: Oligonucleotides involved in Figure 4

Oligo names	Sequence
Oligo1 for α-GAPDH	5'-amine-CATCGCCCTGGACTAGCATACCCATGAACACAAGTTGCGTCACGATGAGACTGGATGAA-3'
Oligo2 for α-GAPDH	5'-OPO_3-TCACGGTAGCATAAGGTGCACGTTACCTTGATTCCCGTCC-amine-3'
Splint	5'-AUAGCUACCGUGAUUCAUCCAGTGAG-3'
Forward Primer	5'-ACCCATGAACACAAGTTGCG-3'
Reverse Primer	5'-GGACGGGAATCAAGGTAACG-3'
Taqman probe	5'-6-FAM-TGGATGAAT/ZEN/CACGGTAGCATAAGGTGCA-IABkFQ-3'